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Do the Austrian blackfly fauna (Diptera: Simuliidae) support the typological approach of the EU water framework directive?

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Abstract

The Austrian blackfly fauna were analysed with regard to spatial (ecoregions, bioregions), vertical (altitude classes) and longitudinal zonation characteristics [stream order, biocoenotic (= fish) regions] on the basis of 2600 investigation sites. Of a total of 45 species recorded, *Simulium ornatum, S. variegatum, S. argyreatum* and *S. reptans* are the most common and most frequently distributed species, occurring in 60% of the investigated sites. Although the Austrian blackfly fauna seem to be quite well documented (on average one investigation site per 32 km^2) the jackknife analyses indicates that there are still some fauna deficits. Whereas the species diversity of the main ecoregions is quite similar, the number of species differs clearly between the bioregions. Within the typological context of the Water Framework Directive, the Austrian Simuliidae confirm the bioregions as the most useful spatial units for river typology. A further optimisation in predicting a target list of blackfly species of a site can be achieved by subdividing the bioregions into either catchment area and altitude classes or into longitudinal zonation types (biocoenotic regions).

Key words: Austria – bioregions – ecoregions – EU water framework directive – longitudinal zonation – rivers – Simuliidae

Introduction

The European Water Framework Directive (WFD) (Council of the European Union 2000) provides the first comprehensive general concept for water management and water protection in Europe. The ecological status of rivers is to be determined on the basis of near-natural reference conditions. The reference-based approach implies, that the ecological status of rivers is to be set up with respect to the near-natural reference conditions. To describe reference conditions, a typological framework is needed. A basic part of this typology is the ecoregion approach, which is explicitly postulated by the WFD.

In contrast to the situation in the United States, where it was possible to invest intensive efforts in the discussion and demarcation of ecoregions (HUGHES & LARSEN 1988; HUGHES et al. 1990; OMERNIK 1987, 1995), the EU states have had comparably little time to develop a similar sophisticated approach. Therefore the member states of the European Union agreed to accept the zoogeographical regions of Europe, compiled by ILLIES (1978), as a basic ecoregion typology. In detail, the Austrian ecoregions have been delineated by a team of experts using a couple of geo-ecological features, quite similar to the delineation process in the US (FINK et al. 2000). Austria, a small landlocked country in Central Europe (with

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an area of about 84,000 km²), has a share of six ecoregions (Fig. 1): the main ones are the Alps (Ecoregion IV), the Central European Mountains (IX), the Hungarian Plains (XI), and the Dinaric Western Balkans (V); the Carpathians (X) and Italy exercise a minor influence (III), (MOOG et al. 2001; SCHMIDT-KLOIBER et al. 2001).

Nevertheless, the ecoregions cover an area that seems to be too big to provide a framework for organising ecological data for a regional management approach. The creation of sub-regions was seen as a fruitful solution and the Austrian aquatic bioregions have been described

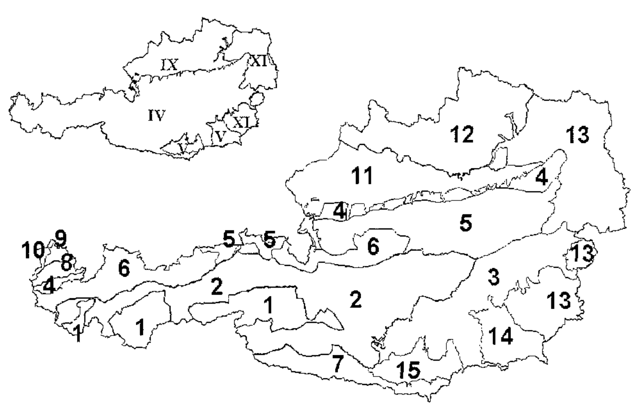

Fig. 1. Aquatic bioregions and ecoregions of Austria. Ecoregions: IV = Alps, V = Dinaric Western Balkans, IX = Central European Mountains, XI = Hungarian Plains; Bioregions: 1 = Glaciated Alps, 2 = Unglaciated Alps, 3 = Ridges & Foothills of the Central Alps, 4 = Flysch, 5 = Limestone Foothills, 6 = Limestone Alps, 7 = Southern Alps, 8 = Helveticum in Vorarlberg, 9 = Alpine Molasse, 10 = Piedmont in Vorarlberg, 11 = Bavarian-Austrian Piedmont, 12 = Austrian Granite & Gneiss Region, 13 = Eastern Ridges & Lowlands, 14 = Grazer Basin & „Grabenland", 15 = Southern Inner-alpine Basins.

by combining the *a priori* approaches and the multivariate a *posteriori* procedures (SCHMIDT-KLOIBER et al. 2002). Based on the ecoregions and the aquatic bioregions, river types can be defined on a national basis using criteria such as altitude, size of the catchment area, stream order etc. (given in Annex II, system A and B, WFD 2000). A map of the Austrian aquatic bioregions is given in Fig. 1. Some characteristic features and the number of sampling sites in a bioregion comprised in this study are provided in Table 1.

A first evaluation of the longitudinal and altitude distribution of Austrian Simuliidae was undertaken by CAR & MOOG (1993) using a smaller data set of about 1150 samples. The aim of this paper is to compare the current status with the 1993 findings and to ascertain whether these ecoregions, bioregions and other typological features might serve as a sound typological basis for the analysis of the blackfly assemblages in evaluating the ecological status of streams within the context of the WFD. On the basis of the species diversity and composition within the spatial context, special emphasis is given to altitude distribution, the effects of the catchment area size and longitudinal zonation patterns with respect to stream order and biocoenotic (fish) regions.

Methods

Data base

The analysis of the Simuliidae fauna is based on the most comprehensive data set available in Austria utilising historical data from earlier studies and current investigations. The data set comprises about 2600 sampling sites distributed over the whole country and adjacent re-

Table 1. Characteristics of bioregions (size of the catchment area, altitude range of sampling sites) and number of research sites.

No.	Bioregion	Area (km²)	Altitude range (m)	No. of sites
1	Glaciated Alps	5163	718–2160	72
2	Unglaciated Alps	15912	348–2017	517
3	Ridges & Foothills of the Central Alps	8265	325–1152	50
4	Flysch	2624	305–978	166
5	Limestone Foothills	9294	300–1146	420
6	Limestone Alps	6290	470–2077	100
7	Southern Alps	2688	405–1110	39
8	Helveticum in Vorarlberg	544	433–880	123
9	Alpine Molasse	287	408–900	132
10	Piedmont in Vorarlberg	207	390–454	101
11	Bavarian-Austrian Piedmont	7277	220–690	582
12	Austrian Granite & Gneiss Region	8637	215–1285	416
13	Eastern Ridges & Lowlands	12194	120–430	391
14	Grazer Basin & „Grabenland"	2331	185–340	16
15	Southern Inner-alpine Basins	1967	367–573	66

gions (Table 1), representing all river-quality categories from near-natural reference conditions and sites of good, moderate, poor and bad ecological status. The data applied are derived from a database that includes information from the Austrian nation-wide surveillance monitoring programme, federal-province monitoring networks, expert reports, applied and basic research studies, dissertations, diploma theses, several studies carried out by the benthic working group of the Department of Hydrobiology, University of Agricultural Sciences, Vienna, as well as the material from the scientific collection of MANFRED CAR (Brunn am Gebirge, Austria). Only data with species-level identification performed or audited by taxonomic experts are included.

The altitude of a sampling site is derived from the digital version of the Austrian Map 1: 50000. The size of the catchment areas is calculated from the tables of the hydrographic maps of Austria (Hydrographischer Dienst 1952–1995). The allocation of the stream order is according to the WIMMER & MOOG (1994) catalogue. The determination of the appropriate biocoenotic region of an investigation site is based on the whole benthic invertebrate community according to the procedure described in the Fauna Aquatica Austriaca (MOOG 1995).

Analysis

As described above, the data set used comprises sources of different origin. Most of the Simuliidae records originate from samples that cover the whole aspect of benthic invertebrates. Some of the data are based on mere Simuliidae samplings. The whole data set - including qualitative and quantitative sampling methods as well as selective selections - was used to describe aspects of the distribution of blackfly species independently of the sampling methods.

The ECOPROF 2.5 software serves as data storage for biotic data and environmental information. The computer program PC-ORD 4.10 for Windows is used for species-area analysis and jackknife estimation of species diversity.

Frequencies listed in Tables 3–6 are calculated as the number of records of a single species in relation to all sites investigated within a class (e.g. stream order 1), expressed in percentages.

Species-area analysis: For species-area analysis using PC-ORD, the whole data set is subsampled randomly 500 times for each research site. If fewer than 2000 subsamples are possible, all possible subsamples are used (McCUNE & MEFFORD 1999).

Jackknife-estimation of species diversity: The number of species observed in a subsample will typically be smaller than the true number of species. PALMER (1990, 1991) compared several ways of estimating the species diversity of an area when it is subsampled with smaller sample units. PC-ORD provides these jackknife estimators of species diversity by assessing the number of species that were missed using non-parametric re-sampling procedure. The "first-order jackknife estimator" (HELTSHE & FORRESTER 1983; PALMER 1990) used is calculated as

$$\text{Jack1} = S + r1(n-1)/n$$

where: S = the observed number of species; r1 = the number of species occurring in one sample unit; n = the number of sample units.

Results

45 Simuliidae species are recorded for Austria (LECHTHALER & CAR 2002; CAR & MOOG 2002). The presence or absence of a species in the four main Austrian ecoregions and 15 bioregions is summarised in Table 2.

32 species (71% of the Austrian blackfly fauna) occur in more than one of the four main Austrian ecoregions. Six species (*Simulium auricoma, S. bertrandi, S. bezzii, S. carpathicum, S. codreanui* and *S. vulgare*) only occur in ecoregion IV, the Alps. Three species (*Twinnia hydroides, S. crenobium* and *S. quasidecolletum*) are found only in ecoregion IX, the Central European Mountains. *S. balcanicum, S. ibariense, S. latipes* and *S. pseudequinum* inhabit only ecoregion XI (Hungarian Lowlands). Of these 13 species that occur only in one ecoregion, 12 are recorded as occurring only in one bioregion.

33 species occur in more than one bioregion. Three of these (*Simulium argyreatum, S. monticola, S. ornatum*) are found in all 15 bioregions, two species (*Prosimulium hirtipes, S. variegatum*) have been recorded in 14, and *S. reptans* in 13 bioregions.

Fig. 2 lists Austrian blackfly species in descending order according to their recorded occurrence. The cumulative frequencies give an impression of the probability of finding a set of certain species at a sampling site and confirms that these six widely distributed species also belong to the most frequently occurring blackflies in Austria. In every second sample, for example, one of the species *S. ornatum, S. variegatum* or *S. argyretum* can be expected. On the other hand, 99.9% of 2600 samples are necessary to find a rare species such as *T. hydroides.*

Table 3 summarises the distribution of Simuliidae species according to the frequency of occurrence in the five altitude categories. The allocation of the altitude of a sampling site to a category is according to the WFD typology given in system A of Annex II. The 500 m line was additionally introduced, because a change in the faunal composition of many Austrian streams takes place in this altitude range, probably owing to the fact that this is where mountain rivers flow into the hilly regions or the plains (BAUERNFEIND & MOOG 2000).

Apart from two species (*S. betrandi, S. crenobium*), it was possible to analyse sufficient data to draw up Table 3. Although Simuliidae occur at all altitudes, it is interesting to note that *P. tomosvaryi* and *S. argyreatum* are the only two species that inhabit rivers across the whole vertical transect.

The frequencies according to the catchment size are presented in Table 4. The ranking of the sampling sites to a size class follows the WFD typology as given in system A of Annex II (2000). The > 2500 km^2 category was introduced to include the Austrian "large rivers" according to the typology of WIMMER et al. (2000). The data set was large enough to provide sufficient analysis for all but four species (*S. bertrandi, S. bezzi, S. crenobium* and *S. latipes*).

The biocoenotic regions and stream orders according to ILLIES & BOTOSANEANU (1963) and STRAHLER (1957) respectively are used to characterise the longitudinal distribution patterns. Because of the limited amount of data from springs, the presentation of the biocoenotic regions starts with the epirhithral (upper trout region) and ends with the metapotamal (bream zone) as no data from littoral and profundal zones exist (Table 5). Methodological details are given in the Fauna Aquatica Austriaca (MOOG 1995) and the Austrian Standards M 6232 (1997).

River sections of stream orders 1 to 7 are treated separately (Table 6). Sections with stream-order 8 and 9 are pooled because there are so few of them (Inn, Morava & Danube). More details on the stream order methodology can be taken from WIMMER & MOOG (1994).

Discussion

State and precision of the documentation

The number of sampling sites differs among bioregions, ranging from 16 to 582 (Table 1). To assess the effect of sampling effort on the number of species found, Simuliid species diversity is compared with the number of sampling sites. Intensively sampled bioregions mostly corre-

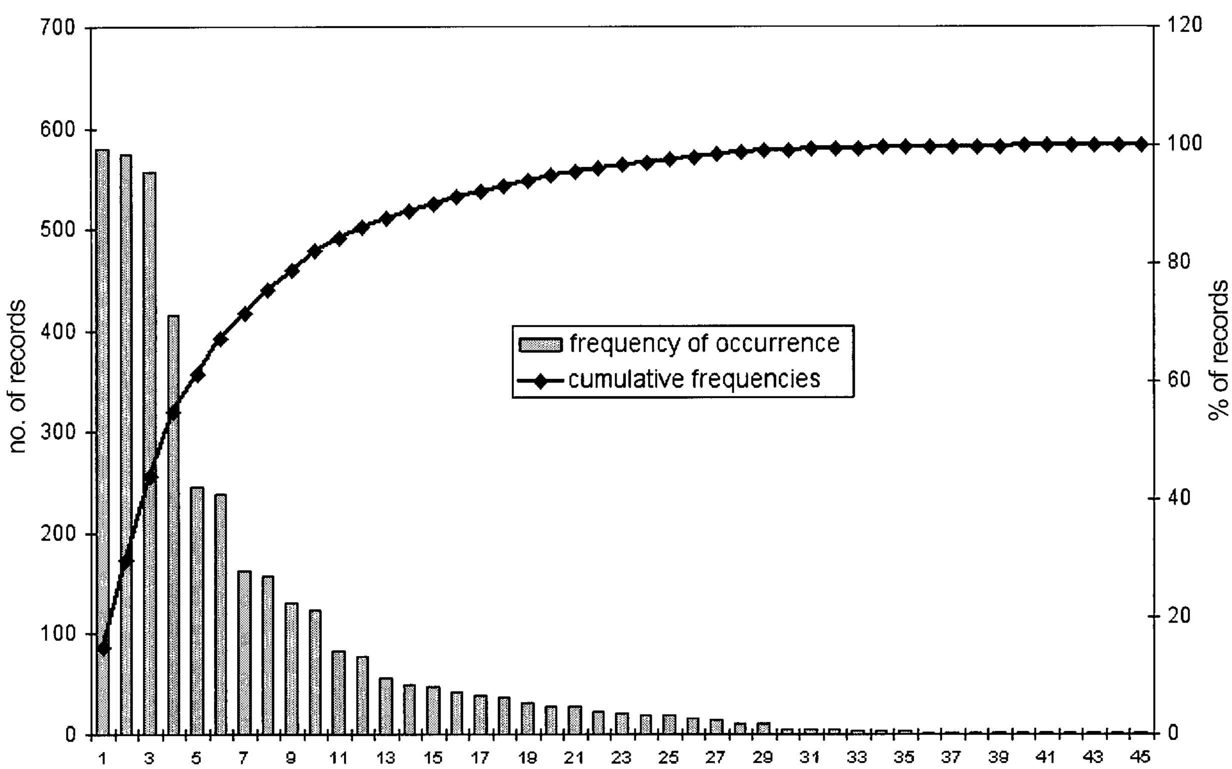

Fig. 2. Frequency curve of blackfly species distribution at sampling sites. 1 = *Simulium ornatum*, 2 = *S. variegatum*, 3 = *S. argyreatum*, 4 = *S. reptans*, 5 = *Prosimulium hirtipes*, 6 = *S. monticola*, 7 = *P. rufipes*, 8 = *S. equinum*, 9 = *S. vernum*, 10 = *S. lineatum*, 11 = *S. argenteostriatum*, 12 = *P. tomosvaryi*, 13 = *S. cryophilum*, 14 = *S. maximum*, 15 = *S. erythrocephalum*, 16 = *S. brevidens*, 17 = *S. trifasciatum*, 18 = *S. aureum*, 19 = *S. costatum*, 20 = *S. angustipes*, 21 = *S. angustitarse*, 22 = *S. noelleri*, 23 = *S. tuberosum*, 24 = *S. intermedium*, 25 = *S. morsitans*, 26 = *S. colombaschense*, 27 = *P. latimucro*, 28 = *S. balcanicum*, 29 = *S. carthusiense*, 30 = *S. degrangei*, 31 = *S. lundstromi*, 32 = *S. pseudequinum*, 33 = *S. posticatum*, 34 = *S. rostratum*, 35 = *S. codreanui*, 36 = *S. bertrandi*, 37 = *S. latipes*, 38 = *S. auricoma*, 39 = *S. bezzi*, 40 = *S. carpathicum*, 41 = *S. crenobium*, 42 = *S. ibariense*, 43 = *S. quasidecolletum*, 44 = *S. vulgare*, 45 = *Twinnia hydroides*.

Table 2. Distribution of Simuliidae species in the Austrian eco- and bioregions (presence/ absence information). For the keys of eco- and bioregion numbers see Fig. 1.

Species	Bioregion															Total no. of bioregions	Total no. of ecoregions
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15		
	Ecoregion																
	IV									IX			XI	V			
Simulium argyreatum	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	15	4
Simulium monticola	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	15	4
Simulium ornatum	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	15	4
Prosimulium hirtipes	1	1	1	1	1	1		1	1	1	1	1	1	1	1	14	4
Simulium variegatum	1	1	1	1	1	1	1	1	1	1	1	1	1		1	14	4
Simulium reptans		1	1	1	1	1		1	1	1	1	1	1	1	1	13	4
Simulium argenteostriatum		1	1	1	1	1	1		1	1	1	1	1		1	12	4
Simulium vernum		1	1	1	1	1		1	1	1	1	1	1	1		12	4
Prosimulium rufipes	1	1		1	1	1	1	1	1	1	1	1				11	2
Simulium cryophilum		1		1	1			1	1	1	1	1	1	1	1	11	4
Prosimulium tomosvaryi		1	1	1	1			1	1		1	1	1			9	3
Simulium trifasciatum		1	1	1	1		1		1		1	1	1			9	3
Simulium equinum		1			1		1			1	1	1	1		1	8	4
Simulium maximum		1	1	1	1	1						1	1			7	3
Simulium tuberosum	1	1			1	1			1		1		1			7	3
Simulium aureum				1	1					1	1	1	1		1	7	4
Simulium erythrocephalum				1	1					1	1	1	1		1	7	4
Simulium lineatum				1	1					1	1	1	1		1	7	4
Simulium angustipes	1			1					1	1	1	1	1			7	3
Simulium angustitarse				1	1					1	1	1	1			6	3
Simulium carthusiense		1		1	1	1					1		1			6	3
Simulium colombaschense			1		1		1				1	1			1	6	3
Simulium costatum		1		1	1					1	1	1				6	2
Simulium brevidens				1	1	1					1	1				5	2
Simulium intermedium								1			1	1	1		1	5	4
Simulium noelleri		1									1	1	1			4	3
Simulium posticatum							1	1				1	1			4	3
Simulium lundstromi										1	1		1		1	4	3
Prosimulium latimucro	1										1	1				3	2
Simulium degrangei		1			1							1				3	3
Simulium morsitans											1	1	1			3	2
Simulium rostratum								1						1	1	3	2
Simulium codreanui		1		1												2	1
Simulium auricoma			1													1	1
Simulium balcanicum													1			1	1
Simulium bertrandi				1												1	1
Simulium bezzi				1												1	1
Simulium carpathicum									1							1	1
Simulium crenobium										1						1	1
Simulium ibariense													1			1	1
Simulium latipes													1			1	1
Simulium pseudequinum													1			1	1
Simulium quasidecolletum												1				1	1
Simulium vulgare					1											1	1
Twinnia hydroides												1				1	1
Total no. within bioregions	9	20	13	24	25	13	10	13	15	19	28	30	29	8	16		
Total no. within ecoregions	36									34			29	16			

Table 3. Distribution and frequencies of occurrence of blackflies according to altitude categories. Frequencies are calculated as the number of records of a single species in relation to all investigated sites within an altitude class, expressed in %.

Species	Altitude class (m)				
	–200 m	200–500 m	500–800 m	800–1500 m	>1500 m
Prosimulium hirtipes	–	5.15	15.16	16.77	6.25
Prosimulium latimucro	–	0.15	0.26	–	12.50
Prosimulium rufipes	–	0.92	4.71	11.72	25.00
Prosimulium tomosvaryi	1.79	3.38	2.88	1.21	1.25
Simulium angustipes	2.24	0.54	0.65	0.20	–
Simulium angustitarse	1.79	0.69	0.26	0.20	–
Simulium argenteostriatum	0.45	2.15	3.79	2.42	–
Simulium argyreatum	15.70	17.06	20.78	22.42	1.25
Simulium aureum	7.62	1.46	0.39	–	–
Simulium auricoma	–	0.08	–	–	–
Simulium balcanicum	4.04	0.15	–	–	–
Simulium bertrandi	no data available				
Simulium bezzii	–	0.08	–	–	–
Simulium brevidens	–	1.31	1.96	0.20	–
Simulium carpathicum	–	–	0.13	–	–
Simulium cartusiense	–	0.31	0.13	–	–
Simulium codreanui	–	–	–	0.40	–
Simulium colombaschense	–	1.08	0.39	–	–
Simulium costatum	0.45	1.77	0.78	0.20	–
Simulium crenobium	no data available				
Simulium cryophilum	1.79	2.23	1.44	0.20	–
Simulium degrangei	–	0.15	0.13	0.61	–
Simulium equinum	15.70	8.76	1.44	0.40	–
Simulium erythrocephalum	5.38	2.38	0.26	–	–
Simulium ibariense	–	0.08	–	–	–
Simulium intermedium	–	1.15	0.65	0.20	–
Simulium latipes	0.45	–	–	–	–
Simulium lineatum	16.59	6.92	0.52	–	–
Simulium lundstromi	0.90	0.38	–	–	–
Simulium maximum	–	1.00	0.39	6.87	–
Simulium monticola	0.45	6.23	12.68	11.52	–
Simulium morsitans	0.90	0.77	0.92	–	–
Simulium noelleri	7.62	0.38	0.13	–	–
Simulium ornatum	19.73	28.21	11.11	3.03	–
Simulium posticatum	–	0.15	0.13	–	–
Simulium pseudequinum	1.35	0.38	–	–	–
Simulium quasidecolletum	–	0.08	–	–	–
Simulium reptans	1.79	19.22	13.20	3.03	–
Simulium rostratum	0.45	0.15	0.13	–	–
Simulium trifasciatum	0.45	1.84	0.92	0.81	–
Simulium tuberosum	–	1.23	1.44	0.40	3.75
Simulium variegatum	1.35	20.14	24.44	12.73	–
Simulium vernum	2.24	3.54	6.80	1.41	–
Simulium vulgare	–	0.08	–	–	–
Twinnia hydroides	–	0.08	–	–	–

late with a comparably high number of species and vice versa. The correlation analysis with an $R^2 = 0.7$ (linear regression) seems to confirm this hypothesis, although some bioregions do not follow this scheme, as documented by the high number of species in the "Flysch" bioregion or the low taxa diversity in the "Unglaciated Alps" [Fig. 3, Flysch (▲) and Unglaciated Alps (■) are highlighted].

When comparing the sampling density at sites investigated with respect to geographically determined areas, not only the number of samples have to be considered,

Table 4. Distribution and frequencies of occurrence of blackflies according to catchment area classes. Frequencies are calculated as the number of records of a single species in relation to all investigated sites within a catchment area class, expressed in %.

Species	Catchment area class (km^2)				
	0–10	10–100	100–1000	1000–2500	>2500
Prosimulium hirtipes	11.31	11.78	11.33	9.87	–
Prosimulium latimucro	1.79	1.92	0.14	0.43	–
Prosimulium rufipes	6.55	10.58	4.97	7.30	–
Prosimulium tomosvaryi	5.36	6.25	3.73	0.43	–
Simulium angustipes	2.38	0.72	0.97	0.43	–
Simulium angustitarse	2.38	1.20	0.55	–	–
Simulium argenteostriatum	–	2.16	5.94	2.58	–
Simulium argyreatum	10.12	21.15	24.86	18.45	–
Simulium aureum	6.55	1.68	0.55	–	–
Simulium auricoma	–	–	–	–	–
Simulium balcanicum	–	–	0.28	–	–
Simulium bertrandi	no data available				
Simulium bezzi	no data available				
Simulium brevidens	0.60	3.85	1.93	–	–
Simulium carpathicum	–	0.24	–	–	–
Simulium carthusiense	–	0.48	0.14	–	–
Simulium codreanui	–	0.24	–	–	–
Simulium colombaschense	–	0.48	1.10	2.15	–
Simulium costatum	11.31	2.16	0.28	–	–
Simulium crenobium					
Simulium cryophilum	9.52	5.05	0.41	–	25.00
Simulium degrangei	–	0.48	0.28	–	–
Simulium equinum	0.60	1.92	8.56	9.44	–
Simulium erythrocephalum	1.79	1.68	1.93	0.86	25.00
Simulium ibariense			3.04		
Simulium intermedium	0.60	1.68	1.24	–	–
Simulium latipes	no data available				
Simulium lineatum	2.38	1.20	7.46	11.59	–
Simulium lundstromi	1.19	0.48	0.28	–	–
Simulium maximum	1.19	2.40	1.80	1.29	–
Simulium monticola	4.17	12.98	11.74	9.87	–
Simulium morsitans	–	0.48	0.55	0.86	–
Simulium noelleri	1.19	0.24	0.41	0.43	–
Simulium ornatum	29.76	25.72	27.76	24.46	50.00
Simulium posticatum	–	–	0.28	0.43	–
Simulium pseudequinum	–	0.24	0.28	–	–
Simulium quasidecolletum	–	–	0.14	–	–
Simulium reptans	2.38	8.89	24.03	19.74	25.00
Simulium rostratum	–	0.24	0.14	–	25.00
Simulium trifasciatum	1.79	3.13	1.93	0.43	–
Simulium tuberosum	0.60	0.96	1.24	2.15	–
Simulium variegatum	7.74	21.63	30.39	33.91	–
Simulium vernum	11.90	12.02	2.90	3.00	–
Simulium vulgare	–	–	0.14	–	–
Twinnia hydroides	–	0.24	–	–	–

but also the different sizes of the bioregions. Consequently, species diversity and sampling density within single bioregions are compared. Fig. 4 shows the correlation between the number of species and the relative number of sampling sites, expressed as the number of sampling sites within an area of 100 km^2. The resulting correlation coefficient (R^2) shows a minimal value of 0.1 (linear regression) indicating hardly any dependency. This provides clear evidence that the number of species is not just a result of a different sampling approach.

Table 5. Distribution and frequencies of occurrence of blackflies according to longitudinal zonation (biocoenotic regions). Frequencies are calculated as the number of records of a single species in relation to all investigated sites within a biocoenotic region, expressed in %.

Species	Biocoenotic region				
	epirhithral	metarhithral	hyporhithral	epipotamal	metapotamal
Prosimulium hirtipes	66.81	58.49	22.29	–	–
Prosimulium latimucro	12.54	1.01	1.68	–	–
Prosimulium rufipes	90.34	23.62	13.06	–	–
Prosimulium tomosvaryi	6.27	22.32	9.52	13.28	–
Simulium angustipes	1.30	2.92	10.71	12.00	–
Simulium angustitarse	–	4.75	6.80	–	–
Simulium argenteostriatum	11.64	17.75	15.10	–	–
Simulium argyreatum	94.71	97.41	75.52	24.41	–
Simulium aureum	1.37	2.40	8.86	87.35	–
Simulium auricoma	no data available				
Simulium balcanicum	–	–	–	24.78	–
Simulium bertrandi	1.29	0.43	–	–	–
Simulium bezzi	no data available				
Simulium brevidens	8.11	12.21	6.61	–	–
Simulium carpathicum	1.33	–	–	–	–
Simulium carthusiense	5.91	1.82	–	–	–
Simulium codreanui	–	0.88	–	–	–
Simulium colombaschense	–	4.59	4.98	–	–
Simulium costatum	3.61	9.84	4.98	–	–
Simulium crenobium	–	0.42	–	–	–
Simulium cryophilum	3.98	16.18	8.65	12.39	–
Simulium degrangei	1.37	0.92	0.84	–	–
Simulium equinum	1.28	11.59	60.05	62.96	349.50
Simulium erythrocephalum	–	1.52	18.09	101.43	349.50
Simulium ibariense	no data available				
Simulium intermedium	–	3.61	7.56	12.65	–
Simulium latipes	no data available				
Simulium lineatum	–	5.08	58.62	198.63	–
Simulium lundstromi	–	–	5.20	–	–
Simulium maximum	6.38	11.14	2.50	–	–
Simulium monticola	64.14	53.55	25.47	–	325.00
Simulium morsitans	–	2.03	2.61	12.33	–
Simulium noelleri	–	0.96	3.47	12.67	–
Simulium ornatum	11.72	106.18	174.98	114.48	–
Simulium posticatum	–	0.91	0.94	12.65	–
Simulium pseudequinum	–	–	0.92	24.78	–
Simulium quasidecolletum	–	0.52	–	–	–
Simulium reptans	19.51	57.50	119.96	98.70	–
Simulium rostratum	–	0.45	1.63	–	–
Simulium trifasciatum	3.80	8.29	9.28	12.24	–
Simulium tuberosum	3.89	4.83	4.91	11.96	–
Simulium variegatum	92.79	126.65	90.35	12.96	–
Simulium vernum	11.44	33.62	21.54	25.04	–
Simulium vulgare	–	–	0.88	–	–
Twinnia hydroides	–	0.50	–	–	–

The data quality of each bioregion was tested on the basis of these conclusions. A sufficient number of sampling sites depends in part on the nature of the bioregion, in particular, on its structural diversity. The species-area curve analysis was applied to examine the adequacy of the number of sampling sites for sufficient documentation of blackfly fauna diversity in a bioregion. The number of species increases with the surface area sampled (number of sampling sites). The degree to which the two variables vary can be measured by how well they correlate.

Table 6. Distribution and frequencies of occurrence of blackflies according to stream order. Frequencies are calculated as the number of records of a single species in relation to all investigated sites within a stream order, expressed in %.

	Stream order							
Species	1	2	3	4	5	6	7	>7
Prosimulium hirtipes	7.00	5.42	5.15	7.63	15.56	9.79	17.91	–
Prosimulium latimucro	1.00	0.49	2.01	0.13	0.14	–	–	–
Prosimulium rufipes	4.00	3.94	4.70	4.87	3.75	6.08	1.49	–
Prosimulium tomosvaryi	2.00	2.96	4.47	3.03	2.02	1.59	–	–
Simulium angustipes	3.00	1.48	–	0.53	0.72	0.53	–	1.16
Simulium angustitarse	–	0.99	1.57	0.39	0.14	0.26	–	–
Simulium argenteostriatum	–	–	1.57	2.11	5.04	2.38	2.24	–
Simulium argyreatum	16.00	7.39	9.17	26.97	20.61	17.72	17.91	4.65
Simulium aureum	14.00	1.48	2.91	0.13	0.43	–	–	4.65
Simulium auricoma	–	–	0.22	–	–	–	–	–
Simulium balcanicum	–	–	–	0.26	–	–	–	10.47
Simulium bertrandi	no data available							
Simulium bezzii	–	–	–	0.13	–	–	–	–
Simulium brevidens	–	0.99	1.34	1.71	1.73	–	–	–
Simulium carpathicum	–	–	–	0.13	–	–	–	–
Simulium carthusiense	1.00	–	0.22	0.26	–	–	–	–
Simulium codreanui	–	–	–	–	0.29	–	–	–
Simulium colombaschense	–	–	–	0.39	0.86	0.79	3.73	–
Simulium costatum	6.00	5.91	2.24	0.26	0.14	–	–	–
Simulium crenobium	no data available							
Simulium cryophilum	7.00	3.45	3.80	1.18	0.29	0.26	1.49	–
Simulium degrangei	–	–	0.22	0.26	0.43	–	–	–
Simulium equinum	1.00	4.93	2.24	5.92	7.35	7.14	6.72	10.47
Simulium erythrocephalum	3.00	0.49	1.12	1.71	0.58	1.32	4.48	8.14
Simulium ibariense	–	–	–	–	–	–	–	–
Simulium intermedium	1.00	0.99	0.67	0.39	1.15	–	1.49	–
Simulium latipes	–	0.99	–	–	–	–	–	–
Simulium lineatum	4.00	–	0.89	1.84	8.65	6.61	–	24.42
Simulium lundstromi	1.00	1.48	0.45	–	0.29	–	–	–
Simulium maximum	–	0.99	1.57	1.05	2.88	3.44	–	–
Simulium monticola	3.00	2.96	6.04	9.34	8.79	14.29	10.45	–
Simulium morsitans	–	2.96	0.45	0.53	0.43	0.53	–	2.33
Simulium noelleri	1.00	0.49	0.22	0.13	0.29	0.26	–	18.60
Simulium ornatum	22.00	17.24	12.98	14.74	22.62	20.63	16.42	13.95
Simulium posticatum	–	–	–	0.13	0.14	–	0.75	–
Simulium pseudequinum	–	–	–	0.53	–	–	–	3.49
Simulium quasidecolletum	–	–	–	–	0.14	–	–	–
Simulium reptans	1.00	4.43	3.13	14.34	18.73	15.34	30.60	1.16
Simulium rostratum	–	–	0.45	–	0.14	–	0.75	–
Simulium trifasciatum	1.00	2.46	1.12	1.18	1.87	0.26	–	–
Simulium tuberosum	1.00	–	0.45	1.32	0.58	0.26	8.21	–
Simulium variegatum	4.00	7.39	6.49	17.11	22.62	31.22	33.58	3.49
Simulium vernum	11.00	6.90	7.61	3.29	1.59	2.91	2.24	–
Simulium vulgare	–	–	–	–	0.14	–	–	–
Twinnia hydroides	–	–	–	0.13	–	–	–	–

The species-area analysis plots the cumulative number of species encountered versus the number of sampling sites, as well as the standard deviations for these values. For the last item, no standard deviation is reported because there is only one possible size: N from a sample of N items. As long as the graph continues to rise, more sites were needed to document the species inventory within an ecosystem completely. The curve levels off when additional sampling sites encounter no new species.

Fig. 5 shows an example of a species-area curve where the total number of species in this bioregion (the Limestone Alps) seems to cover the diversity adequately. The curve increases rapidly at first until it approaches a plateau. The number of species is not expected to increase dramatically with increasing number of sites.

Fig. 6 shows a species-area curve for the Southern Alps (bioregion 7) with an average number of species that is far from approaching a plateau. It is expected that the number of species will significantly increase if the number of sites investigated increases.

Fig. 7 illustrates the difference between observed and jackknife-estimated number of species for all bioregions. The results indicate that some of the bioregions are sampled sufficiently to cover up to 90% (bioregions 4, 5, 6 and 11) of the estimated simuliid species. In contrast to these well-documented regions, the Unglaciated Alps (2), the Southern Alps (7) and the Grazer Basin (14) show a probable species deficit of 35 to 50%. This assumption is confirmed for the Southern Alps by recent research into the adjacent Slovenian Alps (CAR 2001), where 28 simuliid species are reported in an area less

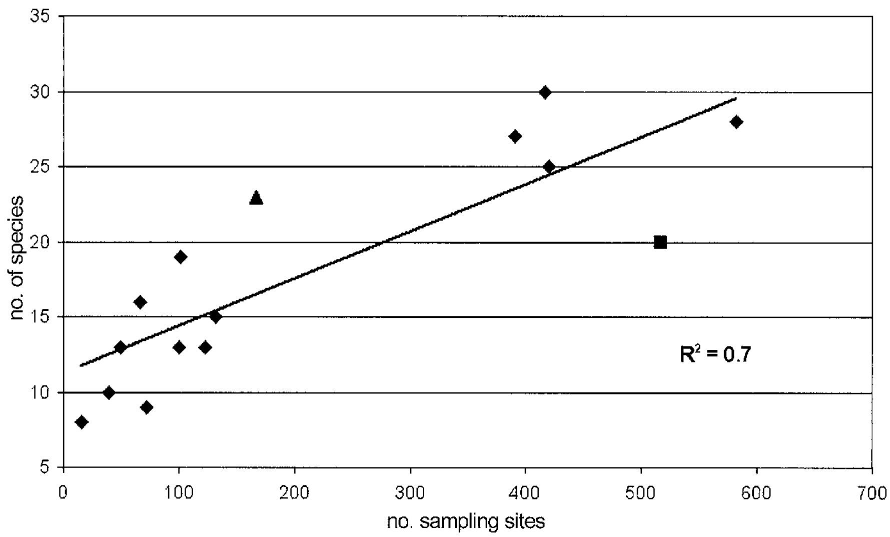

Fig. 3. Correlation between blackfly species richness and number of sampling sites per bioregion (▲ = Flysch, ■ = Unglaciated Alps).

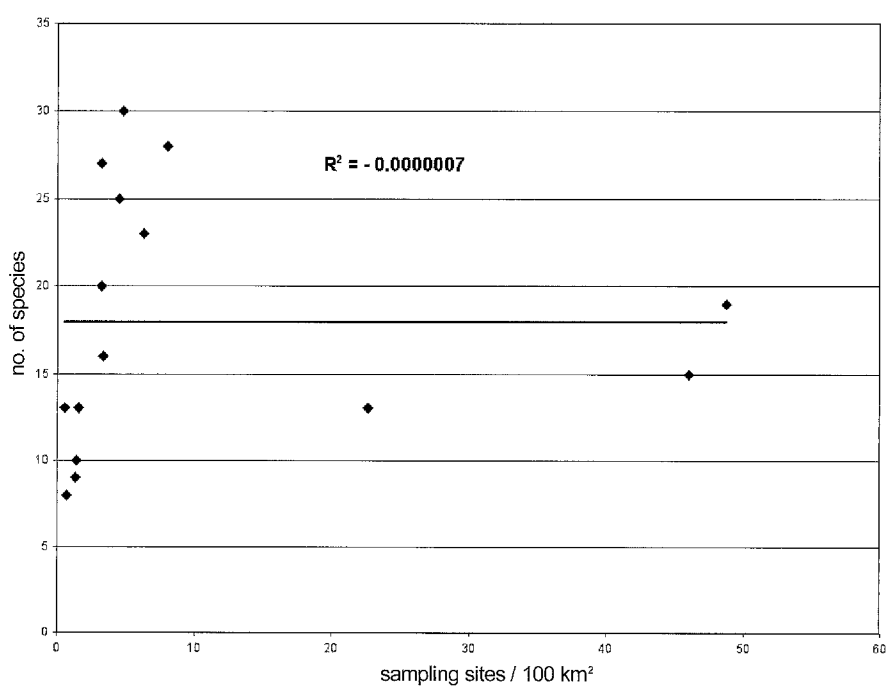

Fig. 4. Correlation between species diversity and the number of sampling sites/100 km^2 within bioregions.

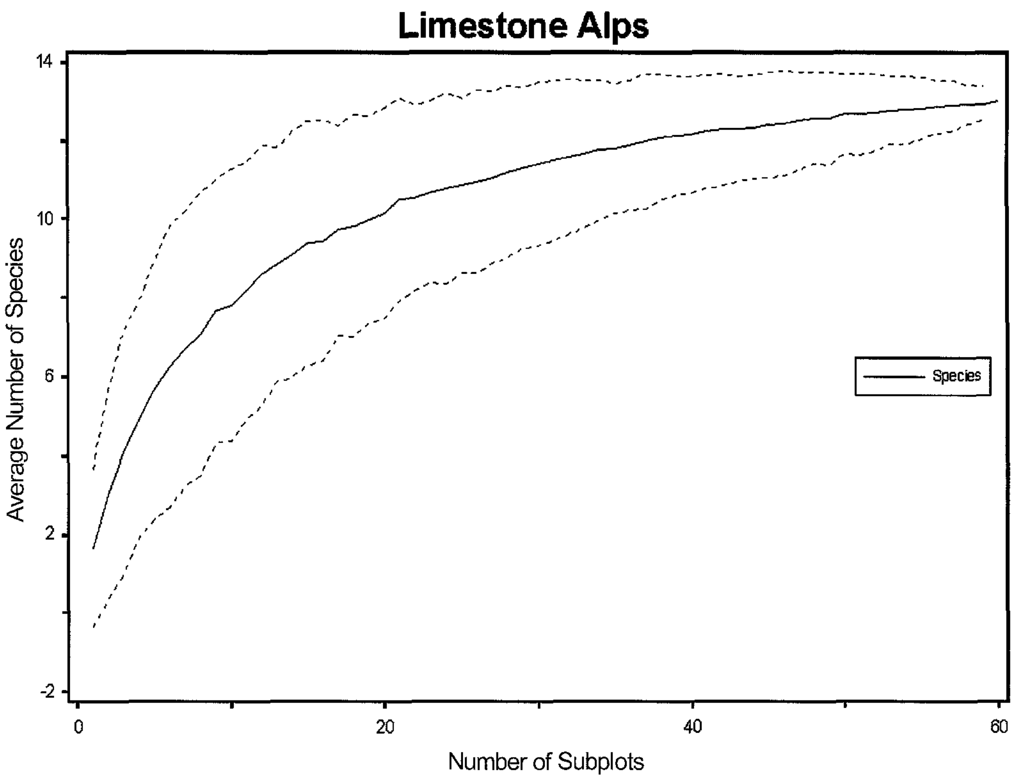

Fig. 5. Species-area curve of Simuliidae records in bioregion 6 (Limestone Alps). Dotted line = standard deviation.

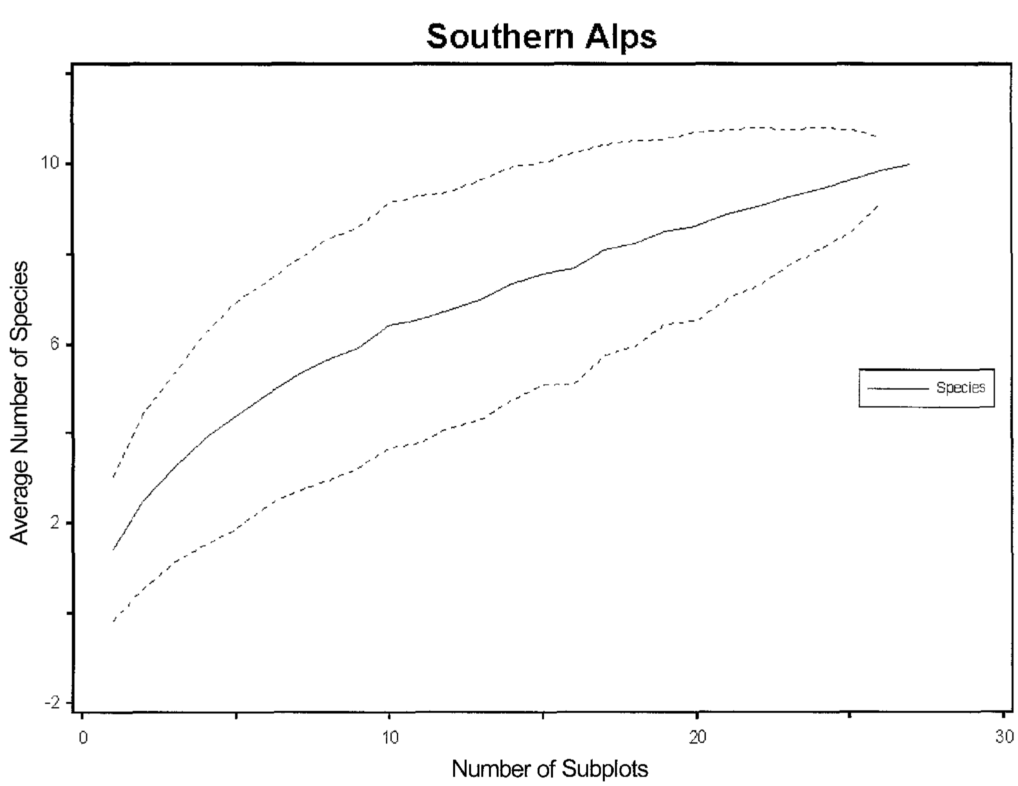

Fig. 6. Species-area curve of Simuliidae records in bioregion 7 (Southern Alps). Dotted line = standard deviation.

than a quarter the size of Austria. So the jackknife estimate of at least 50% missing species of the present amount of 10 species seems to provide reliable results, which can be confirmed by expert opinions.

The jackknife analysis confirms a comparably good status of species documentation for most bioregions, although a probable deficit of some species is also indicated. PALMER (1995) noted that jackknife estimators may not be appropriate when sampling large heterogeneous regions, because the estimated number of species can never be twice the number of observed species. PALMER also warned that these estimators are highly sensitive to the number of rare species observed.

Both possible biases are given in our data set: watercourses – even within a bioregion scale – may differ in many features (size, temperature regime, hydraulic characteristics etc.) and, further, the simuliid fauna include a number of rare species. As a result, the jackknife estimate needs to be seen as a rough estimate, but also as a useful tool to give an indication of whether the number of species investigated is close to reality or far from it.

Simuliidae species diversity and bioregion size

Blackfly biodiversity is defined as the number of different Simuliidae species within an area of interest (ecoregion, bioregion). One of the most fundamental ecological relationships is that the number of different species encountered increases with the size of the region. This species–area relationship may be the oldest ecological pattern recognised. The association of increased area with an increasing number of species at a declining rate has been tested numerous times. It persists over both small and large areas, and with animals as well as plants. Consequently, this hypothesis was tested with respect to the Simuliid fauna of the 15 Austrian bioregions. The H_0 hypothesis was that the number of blackfly species increases with the size of a bioregion.

Fig. 8 indicates that the number of species found in a bioregion correlates only slightly with the bioregion size. The best fitting curve has an R^2-value of only 0.2428. Comparing the area of a bioregion the number of Simuliidae species found, it becomes more clearly evi-

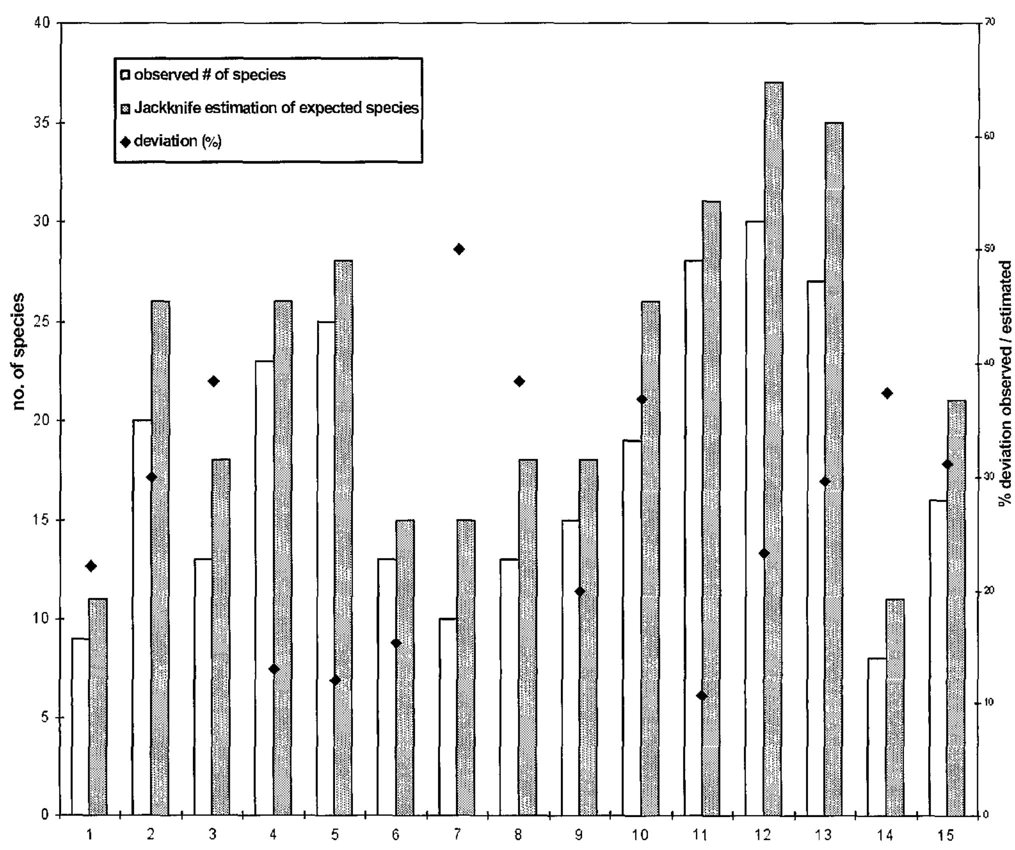

Fig. 7. Observed and estimated blackfly species diversity and % deviation observed.

dent that even very small bioregions are inhabited by numerous species. For example, 19 species have been recorded in the "Piedmont in Vorarlberg" bioregion with an area of only 207 km^2, whereas the large "Ridges and Foothills of the Central Alps" bioregion (8265 km^2) is inhabited by only 13 species. The null hypothesis must be seen as having been falsified: bioregions are inhabited by species numbers independently of their size.

Species diversity and the parameters of the Water Framework Directive (WFD) to characterise stream types

The WFD provides a general definition of the ecological quality of rivers. In this, the composition and abundance of the benthic invertebrate fauna is one of the quality criteria for the classification of the ecological status. A high ecological status is accorded when:

1) Taxonomic composition and abundance correspond completely or nearly completely with undisturbed conditions;

2) the ratio of disturbance sensitive taxa to insensitive taxa shows no signs of alteration from undisturbed levels; and when

3) the level of diversity of invertebrate taxa shows no sign of alteration from undisturbed levels.

Although the term „taxonomic composition" has not so far been defined, many biologists regard the species level as the most important criterion for defining a target list of indicator species of a site. The analysis of the Austrian Simuliidae species is to contribute to this procedure by testing the typological criteria of the WFD.

The number of species within the three largest Austrian ecoregions (Alps, Hungarian Plains, Central European Mountains) is very similar, ranging from 29 to 36. The low number of sites in the rivers of the Dinaric Western Balkans does not permit a comparison of biodiversity. Even when the ecoregions are divided into sub-ecoregions, the number of blackfly species remains similar: 24 species are recorded in the Crystalline Alps (bioregions 1–3) and 26 species are known from the Limestone Alps (bioregions 5–7). In contrast, the taxa diversity in bioregions shows a much greater variation. Species numbers range from 9 to 20 in the three crystalline bioregions and 10 to 25 in the three limestone bioregions. Besides the fact that the composition of species also varies between bioregions this indicates that the use of bioregions as a spatial typological approach will give a better estimate of the expected Simuliid fauna than ecoregions. Consequently, the further analyses focus mainly on taxa diversity of bioregions.

Twelve species (27% of the Austrian blackfly fauna) are recorded in just one bioregion only. Eleven of these species have been found in a few samples (number of findings < 9) and some of them like *S. cordeanui, S. latipes* and *S. ibariense* have recently been recorded for the first time in Austria recently (Car & Lechthaler 2002). These species should be excluded from predictions of target lists until their status as rare or just scarcely found has been more clearly established.

Two species (*S. balcanicum* and *S. pseudequinum*) are common in the Hungarian Plains, an ecoregion identical with bioregion 13 (the Eastern Ridges & Lowlands), a typical habitat for these pannonian species occurring in weedy rivers.

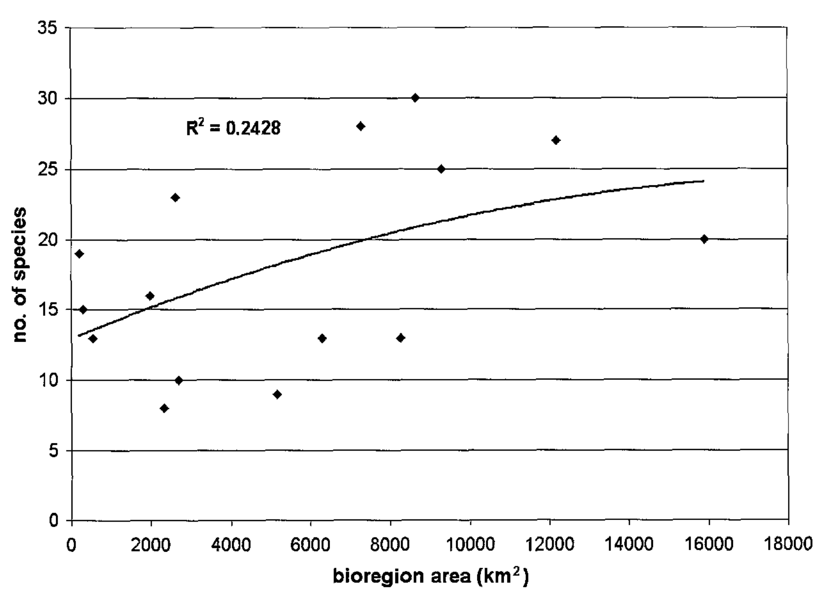

Fig. 8. Correlation of species number vs. size of bioregion area.

Three species (*S. crenobium, S. quasidecolletum* and *T. hydroides*) only occur in ecoregion IX, the Central European Mountains. The most common one, *S. crenobium*, inhabits springs, preferring altitudes of 600 to 1000 m.

Of the six species found only in the Alps, *S. bezzii* has been recorded only once so far, *S. codreanui* twice, the remaining four species occur sporadically.

The vertical zonation of Austrian blackflies

Simuliidae have been found throughout the whole vertical range sampled (130 to 2160 m), but only two species inhabit the complete set of altitude categories: *P. tomosvaryi* and *S. argyreatum* (Table 3). Most of the species (93%) are found between 200 and 500 m, followed by the 500 to 800 m ranges, with 74% of the total species number. The third highest species diversity (25 species) was detected in the rivers below the 200 m line with 56% of the total species (Fig. 9). In the lowland rivers *S. latipes* is the only species that has so far been found only in this altitude range. Twelve species had the highest frequencies below 200 m, which was not to be expected, because the lowlands cover only a small proportion of the Austrian landscape: *S. angustipes, S. angustitarse, S. aureum, S. balcanicum, S. equinum, S. erythrocephalum, S. latipes, S. lineatum, S. lundstromi, S. noelleri, S. pseudequinum* and *S. rostratum*. This group exceeds the maximum frequencies found in all the other altitude classes, as there were nine species between 200 and 500 m, six species between 500 and 800 m, and three species each in the highest frequencies, between 800 and 1500 and above 1500 meters. 51% of the species are found between 800 and 1500 m. Above 1500 m, only six species – 14% of the total – were observed: *P. rufipes, P. latimucro, P. hirtipes, P. tomosvaryi, S. tuberosum* and *S. argyreatum* (in order of the number of findings).

The size of catchment area gives an impression of the development of a river system and serves as an important surrogate parameter to characterise the status of a river. The use of Simuliidae to predict assemblages based on the watershed size may be biased due to the fact that the members of this group show high drift activities. According to the observations of JEDLIČKA et al. (1996), it was expected that rivers with a large catchment area would have a higher number of species by supporting a blackfly population originating from upstream tributaries. This fact cannot be observed in Austria. Large rivers with a catchment area of over 1000 km^2 area show lower species numbers than streams with an area below 1000 km^2.

Only four species colonise rivers of all catchment size categories: *S. erythrocephalum, S. ornatum, S. reptans* and *S. rostratum*. Together with *S. cryophilum*, these four species contribute to the fauna of large rivers with catchment areas over 2500 km^2.

The number of species within the other catchment size classes is significantly higher and ranges from 22 to 36 (Fig. 10). 62.5% of the species can be found in the small catchment areas below 10 km^2. In these streams *S. angustipes, S. angustitarse, S. aureum, S. costatum, S. lundstromi*, and *S. noelleri* are at their maximum frequency, followed by *P. hirtipes, P. latimucro, S. cryophilum* and *S. vernum* which contributed with respect to their second highest frequency. The size catego-

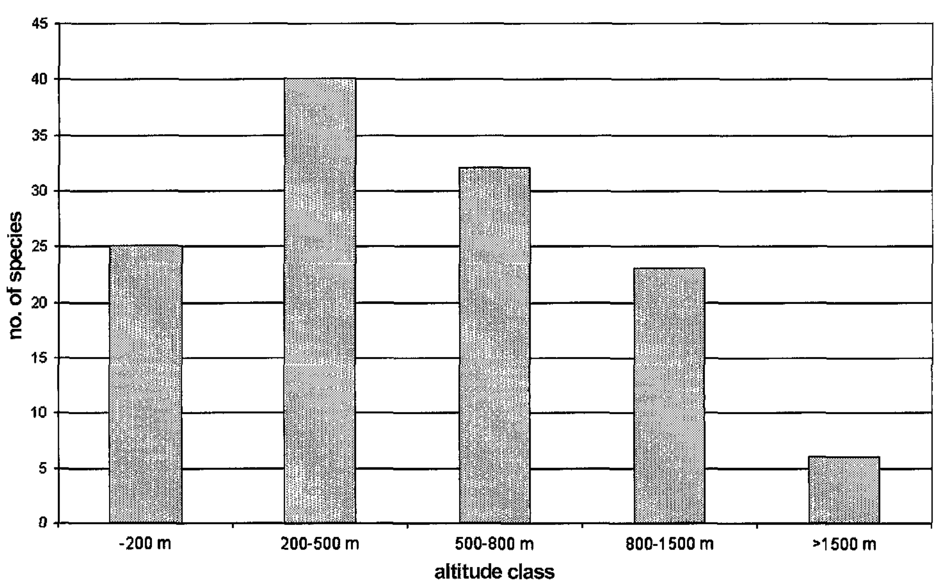

Fig. 9. Species diversity of blackflies along a vertical gradient (altitude classes according to the WFD 2000).

ry between 10 and 100 km² is inhabited by 87.5%, the catchments from 100–1000 km² by 92.5%, those from 1000–2500 by 55%. The highest of the maximum frequency values (15) are found in the catchment between 100–1000 km². All other catchment classes had a highest frequency of between five and seven species.

The analysis of benthic invertebrate **stream zonation** patterns has proved to be a sensitive method of evaluating the ecological status of a community. This method is based on the fact that the distribution of macrobenthic organisms changes along the longitudinal course of a river, as serial trends in hydro-morphological and physical-chemical conditions change. This phenomena has been observed for almost 140 years, and in Europe it has led to a subdivision of rivers into typical fish regions that have been transferred into the concept of biocoenotic regions (the Rhithron-Potamon Concept) by ILLIES & BOTOSANEANU (1963).

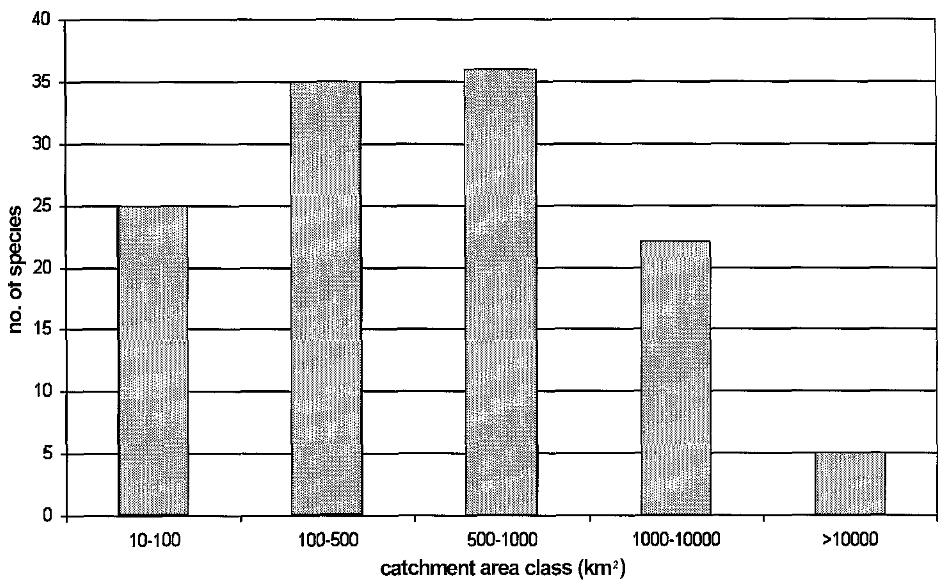

Fig. 10. Species diversity vs. size of catchment area.

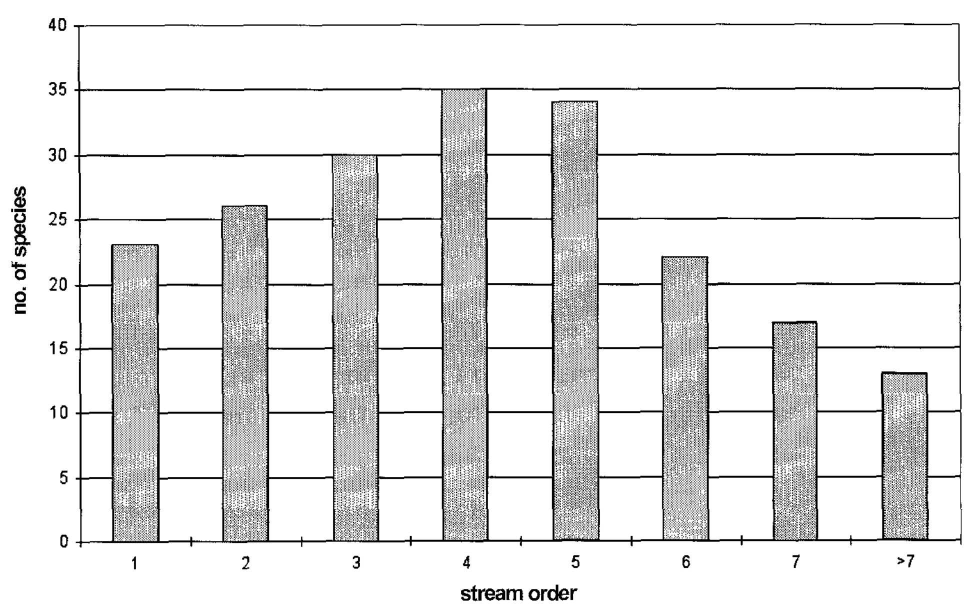

Fig. 11. Species diversity vs. stream order.

The compliance with, or deviation from this state can be evaluated by comparing the zonation structure of a reference stretch or a type specific target community with the actual fauna of an investigated site. Using the Rhithron-Potamon concept, the Austrian Standards M 6232 (1997) describe the degrees of deviation for evaluating the ecological functionality of a river section by means of the longitudinal zonation structure.

13 species each colonise sections of three or four adjacent biocoenotic regions. Six species are to be found only in two, and eight species in only one biocoenotic region (Table 5). Only one species, *Simulium monticola*, inhabits rivers in all five biocoenotic regions. The upper trout zone (epirhithral) is colonised by 24, the lower trout zone (metarhithral) by 36, the grayling zone (hyporhithral) by 33, and the barbel zone (epipotamal) by 20 blackfly species. The metapotamal (bream zone) is inhabited by only three species: *S. equinum*, *S. erythrocephalum*, and *S. monticola.* All three of them are widely distributed in all river types and cannot be regarded as typical inhabitants of the bream zone. The highest number – 17 maximum frequencies – is observed in the grayling zone, followed by the metarhithral (13), epipotamal (11), and epirhithral (7).

Stream orders are used as a simple but effective tool for classifying running waters. They are seen as surrogates taking in account a couple of physical and hydromorphological features changing from source to the mouth of a running water system. For Austrian rivers the Strahler-stream orders have been correlated with river length, width, catchment area, mean run-off, and other aspects (Wimmer & Moog 1994).

Most species can be found along comparatively long sections of adjacent stream orders, but – probably due to sampling deficits – are not continuously recorded. If within a consistent series of documentation single in-between stream orders with zero records are not registered, 10 species can be found in all stream orders, and nine species in seven stream orders. Together, this gives more than 40% of the species, which are distributed over a fairly wide range of stream orders. 17 species inhabit four to six stream orders with a maximum of nine species covering five stream orders. Owing to their scarcity, eight species can be found only in one stream order.

Compared to Car & Moog (1993) there has been a change in the species number per stream order (Fig. 11). The currently observed decrease of species in stream order 1, from 32 to 23, is caused by a new ranking of the Teichbach in Lunz, which has the highest species diversity recorded in Austria (16 species). As this brook is now recognised as a side branch of the Upper Lunzer Seebach, its stream order has changed from 1 to 2. The number of species stream order 2 has consequently increased from 21 to 26. As most samples originated from rivers of stream orders 3 to 6, their numbers of species have increased (order 3: from 17 to 30; order 4: from 23 to 35; order 5: from 20 to 34; order 6: from 12 to 22). A smaller increase was observed in stream order 7 (from 12 to 17) and stream orders > 7 (from 7 to 13). The striking increase of species between stream order 3 and 4 observed in 1993, which according to Statzner & Higler (1986) was related to changes in the hydraulic stress, can no longer be observed, although the 4^{th} order rivers still have the highest number of blackfly species (Fig. 11).

Conclusion and Outlook

Austrian blackfly fauna are well documented, although it is statistically evident that, even after 26 years of intensive sampling, some of the Austrian blackfly fauna still remain unknown. The jackknife analysis visualises the sampling deficits and provides two opportunities: 1) to have an opinion about the reliability of the current data, and 2) to design a sampling programme to fill in the "blank spaces" on the distribution map.

Based on the blackfly assemblages – as if a substitute of the benthic invertebrate community of rivers – the WFD's typological approach can optimally operate on different levels. The primary level is the area-based geographic framework and includes ecoregions, which need to be broken down to bioregions. The secondary level needs to differentiate the bioregion into either catchment areas and altitude categories or into longitudinal zonation types (biocoenotic regions). The study clearly shows that, within this secondary level, a set of typical features can be pointed out.

The first group are the four to six species that are ubiquitous and occur at almost every site: *Simulium ornatum, S. variegatum, S. argyreatum, S. reptans, Prosimulium hirtipes,* and *S. monticola.* The second group are the species that colonise stream or river sections of a certain type (e. g. headwaters, lowland rivers). The third group consists of the species that have a distinct maximum distribution that can be related to environmental features such as altitude categories, catchment areas, stream orders or biocoenotic regions.

By putting all this information together, a target list of possibly occurring blackfly species can be drawn up.

This analysis of the Austrian blackfly fauna does not solve the problems associated with the implementation of the European Water Framework Directive, especially focusing on the evaluation of the ecological status of a water body. But it contributes to solving the problem of defining target lists by pointing out the most useful typological features as bioregions, broken down according to features of altitude and catchment size or biocoenotic regions.

Acknowledgements

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Zusammenfassung

Anhand von etwa 2600 Untersuchungsstellen wird die Verbreitung der österreichischen Simuliidenfauna im Sinne der typologischen Vorgaben der EU-Wasserrahmenrichtlinie (WRRL) ausgewertet. Es werden ausschließlich qualitätsgesicherte Daten mit Determination auf Artniveau verwendet. Die räumlichen Typkriterien umfassen die Ökoregionen und die Fließgewässer-Bioregionen, als weitere Charakteristika werden Seehöhenklassen, Einzugsgebietsgröße, Flussordnungszahl und biozönotische Region einer Probenstelle herangezogen. Von 45 nachgewiesenen Arten kommen *Simulium ornatum, S. variegatum,* und *S. reptans* an 60% der Sammelstellen vor. Obwohl die österreichische Simuliidenfauna gut dokumentiert ist (durchschnittlich 1 Untersuchungsstelle auf 32 km^2), zeigt die Jackknife-Analyse, dass immer noch Sammeldefizite vorhanden sind. Während in den Ökoregionen die Artenzahlen sehr ähnlich sind, unterscheiden sich die Bioregionen in ihrem Artenreichtum deutlich. Innerhalb des typologischen Kontexts der Wasserrahmenrichtlinie erweisen sich die Bioregionen als die aussagekräftigsten räumlichen Einheiten für eine Fließgewässertypologie. Eine weitere Unterteilung der typologischen Betrachtungsebene in Seehöhenklassen und Einzugsgebiete oder biozönotische Region unterstützt die Ausweisung eines Leitbildes für Referenzstrecken hinsichtlich der taxonomischen Zusammensetzung der Simuliidenfauna (entspricht den Systemen A und B in Anhang II der WRRL, 2000).

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